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Karyology of Five Species of Bats (Vespertilionidae, Hipposideridae, and Nycteridae) from Gabon with Comments on the Taxonomy of *Glauconycteris*

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Abstract

We karyotyped five species of bats from Gabon. *Glauconycteris beatrix* and *G. poensis* both have an all-biarmed 2n = 22 karyotype, consistent with the recognition of *Glauconycteris* as a genus distinct from *Chalinolobus*. One specimen of *Hipposideros caffer* had a 2n = 32 karyotype similar to that published for this species from other areas in Africa. We report a 2n = 52 karyotype for *Hipposideros gigas* which is identical to that found in *H. vittatus*. The slit-faced bat *Nycteris grandis* has a 2n = 42 karyotype similar to that known in other species of *Nycteris*.

Key words: chromosomes, Gabon, *Glauconycteris*, *Hipposideros*, karyotypes, *Nycteris*, Rabi, taxonomy

Introduction

The Republic of Gabon includes extensive tracts of tropical rain forest and has an economy based largely on oil production. A recent study of biodiversity (Alonso et al. 2006; Lee et al. 2006) focused on the Rabi Oilfield, which is located in the Gamba Complex of Protected Areas in the Ogooué-Maritime Province of southwestern Gabon. This study included a survey of small mammals during February and March of 2002 (O'Brien et al. 2006; Rodriguez et al. 2006) that documented the presence of 13 chiropteran species in the rainforest of the Rabi Oilfield. Primus et al. (2006) reported karyotypes of four species of shrews, seven species of rodents, and five species of megachiropteran bats collected at Rabi. However, they did not describe chromosomal data for the microchiropteran specimens pending confirmation of species identifications. We report here karyotypes for five species of microchiropteran bats representing three families.

MATERIALS AND METHODS

Bats were collected in the Rabi Oilfield by mist netting as described by Rodriguez et al. (2006). Specimens examined for karyotypic data are listed in the Appendix with specific collecting localities. Chromosome slides and cell suspensions of bone marrow were prepared in the field using the methods of Baker et al. (2003) as modified by Primus et al. (2006). Standard geimsa-stained karyotypes were prepared from these blaze-dried slides.

RESULTS

Glauconycteris.—We karyotyped a single specimen each of the Beatrix butterfly bat (*G. beatrix*) and the Abo butterfly bat (*G. poensis*) and found that both of these vespertilionid species had a diploid number of 22 with all chromosomes being distinctly biarmed (Fig. 1A-B). In both species, ten pairs are metacentric or slightly submetacentric and one pair (the smallest) is subtelocentric. The sex chromosomes could not be identified with certainty in either species.

Hipposideros.—We karyotyped two species of the hipposiderid genus *Hipposideros*. One male specimen of Sundevall's leaf-nosed bat (*H. caffer*) had a 2n = 32 karyotype with all chromosomes being biarmed (Fig. 1C). A single male of the giant leaf-nosed bat (*H. gigas*) had a diploid number of 52 with 6 pairs of biarmed and 20 pairs of acrocentric chromosomes (Fig. 1D).

Nycteris grandis.—One male of this nycterid species was karyotyped, and we found a diploid number of 42, with 34 biarmed and 6 acrocentric autosomes (Fig. 1E). The X is a large subtelocentric and the Y is a small acrocentric.

DISCUSSION

Chromosomal systematics of Glauconycteris.—We examined two species of the vespertilionid genus *Glauconycteris*. Bickham (1979a, b) identified the karyotype of *Myotis nigricans* (2n = 44) as primitive for the family. This primitive vespertilionid karyotype includes an autosomal complement of four biarmed and 17 acrocentric pairs. In the primitive vespertilionid karyotype, eight autosomal arms are fused to form four biarmed chromosomes. Routenbach et al. (1993) described (but did not illustrate) a 2n = 18 all-biarmed karyotype for *Glauconycteris variegata*. Routenbach et al. (1993) reported two pairs of metacentric, five pairs of submetacentric, and one pair of subtelocentric autosomes in *G. variegata*.

Our karyotypes for *G. beatrix* and *G. poensis* (Fig. 1A-B) both show a diploid number of 22, with all chromosomes biarmed. The karyotypes of the two species differ in the length of the short arms of the two smallest chromosomes. Volleth and Heller (2007) reported a similar karyotype for *G. beatrix* from the Democratic Republic of Congo. Our karyotype of *G. beatrix* from Gabon (Fig. 1A) differs from Volleth and Heller's (2007) specimen in that the smallest chromosome pair was subtelocentric in our specimen, rather than submetacentric. Also, relative to other chromosomes, this chromosome pair appears distinctly smaller in our Gabonese specimen than in the bat examined by Volleth and Heller (2007). Volleth and Heller (2007) reported extensive regions of paracentromeric heterochromatin in *G. beatrix*, and it seems likely that the difference in size and morphology of the smallest chromosome is due to differing amounts of heterochromatin in the small arm. The variation could be attributed to polymorphism, geographic variation, or to some other more complex taxonomic issues. Population studies and more extensive geographic sampling would be required to resolve the question.

The sex chromosomes of *G. variegata* consist of a medium-sized subtelocentric X and a small metacentric Y (Routenbach et al. 1993). The male karyotype

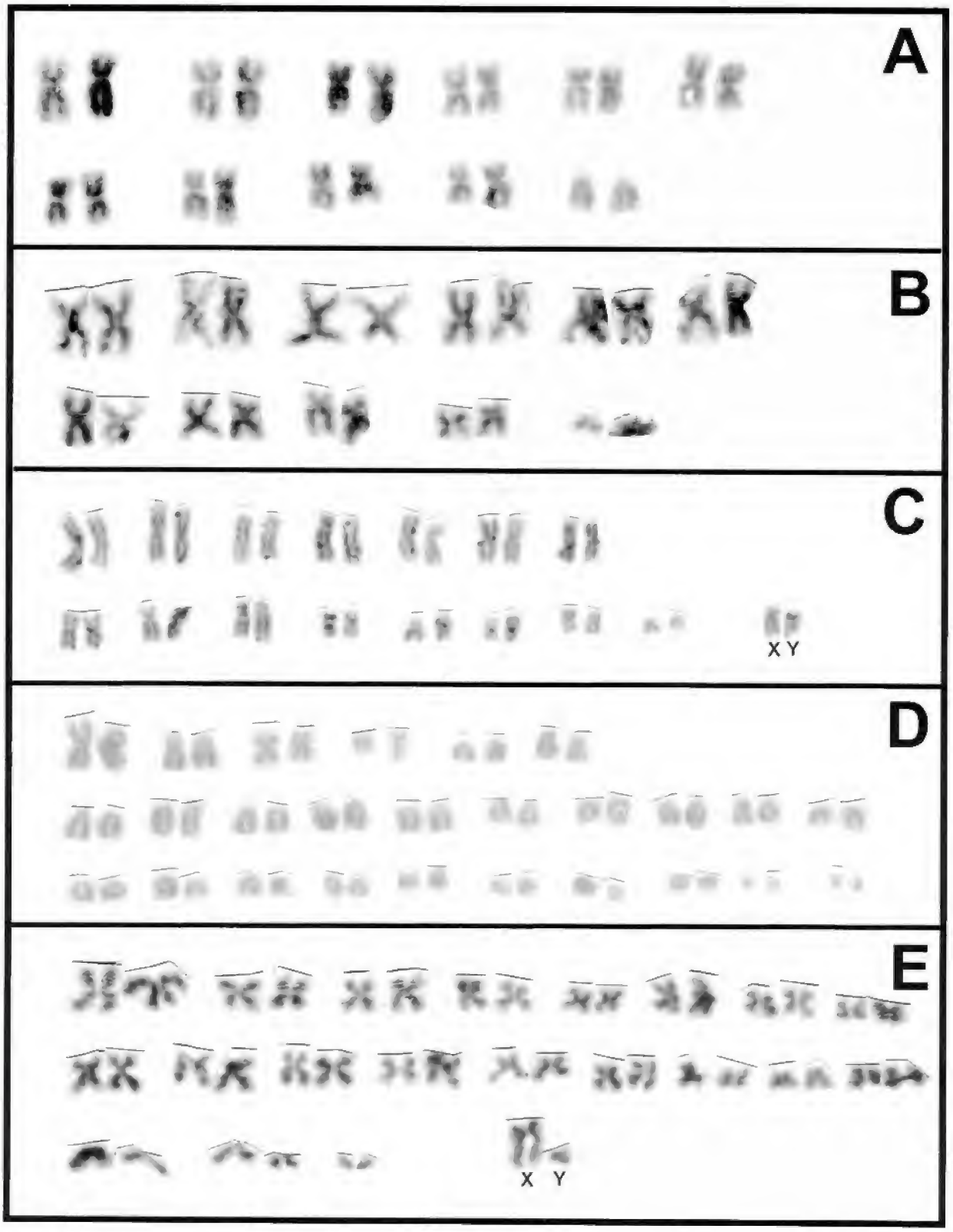

Figure 1. Standard karyotypes of bats collected at the Rabi Oilfield, Gabon. A. *Glauconycteris beatrix* female TK110482; B. *Glauconycteris poensis* male TK110464; C. *Hipposideros caffer* male TK110388; D. *Hipposideros gigas* male TK110279; E. *Nycteris grandis* male TK110435.

of *G. beatrix* has not been described, but Volleth and Heller (2007) reported submetacentric X chromosomes in *G. beatrix*, with the short arm being an autosomal element fused to the sex chromosome. Based on chromosomal morphology, we were not able to identify sex chromosomes in either species of *Glauconycteris* (Fig. 1A-B). There is not an obvious heteromorphic pair in the male *G. poensis* (Fig. 1B). The similar karyotypes of *G. poensis* and *G. beatrix*, combined with the lack of chromosomal heteromorphism in the male *G. poensis*, suggests a neo-XY sex-determining system in these species. Heterochromatic additions (such as documented by Volleth and Heller, 2007) may be important in chromosomal evolution of *Glauconycteris*, and it is therefore plausible that the lack of sex chromosome heteromorphism in *G. poensis* is due to heterochromatic additions to the Y chromosome. The overall similarity of the karyotypes of the female *G. beatrix* and the male *G. poensis* lends credence to the hypothesis that both the X and the Y chromosomes have fused to an autosome in these species.

Hoofer and Van Den Bussche (2003) examined four species of *Glauconycteris* and their phylogeny shows *G. beatrix* and *G. variegata* to be sister taxa, with *G. poensis* being sister to *G. argentatus*. Given this phylogeny, it is plausible that the karyotype of *G. poensis* and *G. beatrix* is primitive for the genus, while the 2n = 18 karyotype is derived in *G. variegata*.

This genus has been traditionally associated with *Chalinolobus* and until recently, *Glauconycteris* was considered a subgenus of *Chalinolobus* (Ryan 1966; Koopman 1971; Aggundey and Schlitter 1984). However, molecular data (Hoofer and Van Den Bussche 2003) and studies of bacular morphology (Hill and Harrison 1987) have provided strong evidence against this association. Recent authorities (Nowak 1999; Eger and Schlitter 2001; Hoofer and Van Den Bussche 2003; Simmons 2005) regard *Glauconyteris* as a distinct genus. Volleth and Tidemann (1989) reported a 2n = 44 karyotype in *Chalinolobus morio*. The significant difference in karyotype supports the contention (Eger and Schlitter 2001; Hoofer and Van Den Bussche 2003) that *Glauconycteris* is unrelated to *Chalinolobus*.

Karyotypes of Hipposideros.—Our 2n = 32 karyotype (Fig. 1C) for *H. caffer* is similar to that previously reported for the species by Đulić and Mutere (1974), Peterson and Nagorsen (1975), and Rautenbach et al. (1993), but the putative Y chromosome is biarmed, rather than acrocentric as was described for this species by Peterson and Nagorsen (1975) and Rautenbach et al. (1993). The karyotype of *H. caffer* is similar to most other congeneric species which have been published (Hood et al. 1988; Rautenbach et al. 1993; Sreepada et al. 1993), with the exception of *H. commersoni* (Rautenbach et al. 1993) and *H. gigas* (this study).

Hipposideros gigas has been regarded by some authorities as a subspecies of *H. commersoni*, but Simmons (2005) recognizes this taxon (along with *H. vittatus* and *H. thomensis*) as a distinct species from *H. commersoni*. The diploid number of 52 we found in our Gabonese specimen of *H. gigas* (Fig. 1D) is identical to that reported by Rautenbach et al. (1993) for five South African specimens identified as *H. commersoni*. Based on locality, the Routenbach et al. (1993) specimens are likely representatives of the taxon now recognized (Simmons 2005) as *H. vittatus*. Routenbach et al. (1993) regarded the largely acrocentric karyotype of their South African specimens as derived from the typical biarmed (2n = 32) *Hipposideros* karyotype (Fig. 1C) by a series of centric fissions. It appears that the derived 2n = 52 karyotype is found in both *H. gigas* and *H. vittatus*, and it seems likely that a similar karyotype would be found in other species of the *commersoni* group (Simmons 2005).

Karyology of Nycteris.—The 2n = 42 karyotype of *N. thebaica* was first reported from Zimbabwean specimens by Peterson and Nagorsen (1975) and confirmed by Rautenbach et al. (1993). Both *N. woodi* (Rautenbach et al. 1993) and *Nycteris hispida* (Lee et al. 1989) have a similar 2n = 42 karyotype. All three species have 19 pairs of biarmed autosomes and 1 pair of acrocentric autosomes. *N. woodi* has only four pairs of metacentric autosomes, and 12 submetacentric. The X is metacentric, with a small acrocentric Y. *N. thebaica* differs in having nine metacentric, seven submetacentric, and three subtelocentric pairs among the autosomes. In *N. thebaica*, the X is submetacentric and the Y is a small metacentric.

The karyotype of *N. macrotis* (Lee et al. 1989) differs from those previously discussed in having a diploid number of 40 and four fewer autosomal arms. The sex chromosomes of *N. macrotis* are both metacentric.

Nycteris grandis is a member of the *hispida* group (Koopman 1975). Our specimen of *N. grandis* resembles most other species of *Nycteris* in having a diploid number of 42, but with 17 biarmed and three acrocentric autosomal pairs. The X is a large subtelocentric, and the Y is a small acrocentric (Fig. 1E). We did not see secondary constrictions such as those found by Peterson and Nagorsen (1975) in *N. thebaica*, and no constrictions were reported by Rautenbach et al. (1993) in the 18 specimens of *Nycteris* they examined from Namibia and South Africa.

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APPENDIX

Specimens examined for chromosomal data. TK = Texas Tech University voucher number. Tissues and chromosome preparations are archived at Texas Tech University under the TK number. NMNH = voucher specimen at the Smithsonian Institution. All specimens are standard skin, skull, and skeleton preparations.

GABON: Ogooué-Maritime Province; Rabi Oilfield, 1 km northeast of Well 45, UTM 32M-9793059N-596735E: *Glauconycteris beatrix* NMNH584723, TK110482, female; *Glauconycteris poensis* NMNH584724, TK110464, male; *Hipposideros gigas* NMNH584720, TK110279, male; *Nycteris grandis* NMNH584722, TK110435, male. 100 m north of Well 59, UTM 32M-9784842N-594824E: *Hipposideros caffer* NMNH 584718, TK110388, male.

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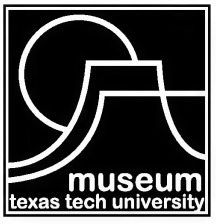

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